Mammalian Biology 92 (2018) 68–77

Contents lists available at ScienceDirect

Mammalian Biology

journal homepage: www.elsevier.com/locate/mambio

Original Investigation

Genetic polymorphism and structure of wild and zoo populations of the fosa (Eupleridae, Carnivora), the largest living carnivoran of Madagascar

Géraldine Veron[a,*], Délia Dupré[a], Mia-Lana Lührs[b], Peter M. Kappeler[b], Luke Dollar[c,d], Julie Pomerantz[d], Steven M. Goodman[e,f]

[a] Institut de Systématique, Evolution, Biodiversité (ISYEB), Muséum national d'Histoire naturelle, CNRS, Sorbonne Université, EPHE, CP 51, 57 rue Cuvier, 75231, Paris Cedex 05, France
[b] Department of Sociobiology/Anthropology, Johann-Friedrich-Blumenbach-Institute of Zoology and Anthropology, Georg-August-University, Kellnerweg 6, 37077, Göttingen, Germany
[c] Nicholas School of the Environment, Duke University, Durham, NC, 27708, USA
[d] Department of Environmental Sciences and Sustainability, Catawba College, Salisbury, NC, 28144, USA
[e] Field Museum of Natural History, 1400 South Lake Shore Drive, Chicago, IL, 60605, USA
[f] Association Vahatra, BP 3972, Antananarivo 101, Madagascar

ARTICLE INFO

Article history:
Received 30 November 2017
Accepted 11 April 2018
Available online 14 April 2018

Handled by Paul Grobler

Keywords:
Conservation
Cryptoprocta ferox
Genetics
Phylogeography
Studbook

ABSTRACT

Cryptoprocta ferox, or fosa, is the largest living endemic carnivoran of Madagascar, with presumably high dispersal capacity, and for which no broad scale phylogeographic study has been conducted to date. This species is considered "Vulnerable" by the IUCN and the subject of a captive breeding program; approximately 113 individuals are held in 57 zoos. The aim of this study was to examine the genetic structure and polymorphism within both captive and wild populations, to determine possible lineage variation, and to make recommendations for the captive breeding program. For this purpose, we analyzed three mitochondrial (Cytochrome b, ND2, Control Region) and one nuclear (Beta-fibrinogen intron 7) markers. The results showed an overall low level of genetic polymorphism, likely related to its dispersal capacity, and some genetic structure possibly associated with geographical barriers, such as large rivers. The genetic diversity of the captive population was greater than that of wild individuals included herein, suggesting that the captive population encompasses a considerable proportion of the genetic diversity of the species. This genetic variability is presumably the consequence of frequent imports of wild animals into zoos from different areas of Madagascar, and subsequent exchanges between zoos. Based on the low overall genetic polymorphism of the species and the absence of deeply divergent lineages, we recommend the continued mixing of captive animals. Our results may help the management of the fosa in the wild and in captivity, which is crucial for a species that faces many threats in the wild, in particular habitat degradation and hunting pressure. In any case, enhanced protection of the species and its forested habitat is urgently needed.

Introduction

Knowledge of population genetic structure for species under threat is crucial, allowing for the identification of conservation or management units (Manel et al., 2003). Dispersal capacity is known to have a major influence on population genetic structure and gene flow (Cushman and Lewis, 2010). Home range area, geographic range, and body mass are the most important predictors of dispersal capacity in mammals (Whitmee and Orme, 2013).

Cryptoprocta ferox Bennett, 1833 (family Eupleridae), also known by the vernacular name fosa or fossa, is a solitary carnivoran endemic to Madagascar. The Malagasy carnivorans (Eupleridae) separated 18–24 Mya from their closest relatives, the Herpestidae (mongooses); within the Eupleridae, the fosa was found to be either

* Corresponding author.
E-mail addresses: geraldine.veron@mnhn.fr (G. Veron), delia.dupre@outlook.fr (D. Dupré), mluehrs@gwdg.de (M.-L. Lührs), pkappel@gwdg.de (P.M. Kappeler), luke@duke.edu, luke.dollar@catawba.edu (L. Dollar), juliepomerantz@gmail.com (J. Pomerantz), sgoodman@fieldmuseum.org (S.M. Goodman).

https://doi.org/10.1016/j.mambio.2018.04.007
1616-5047/

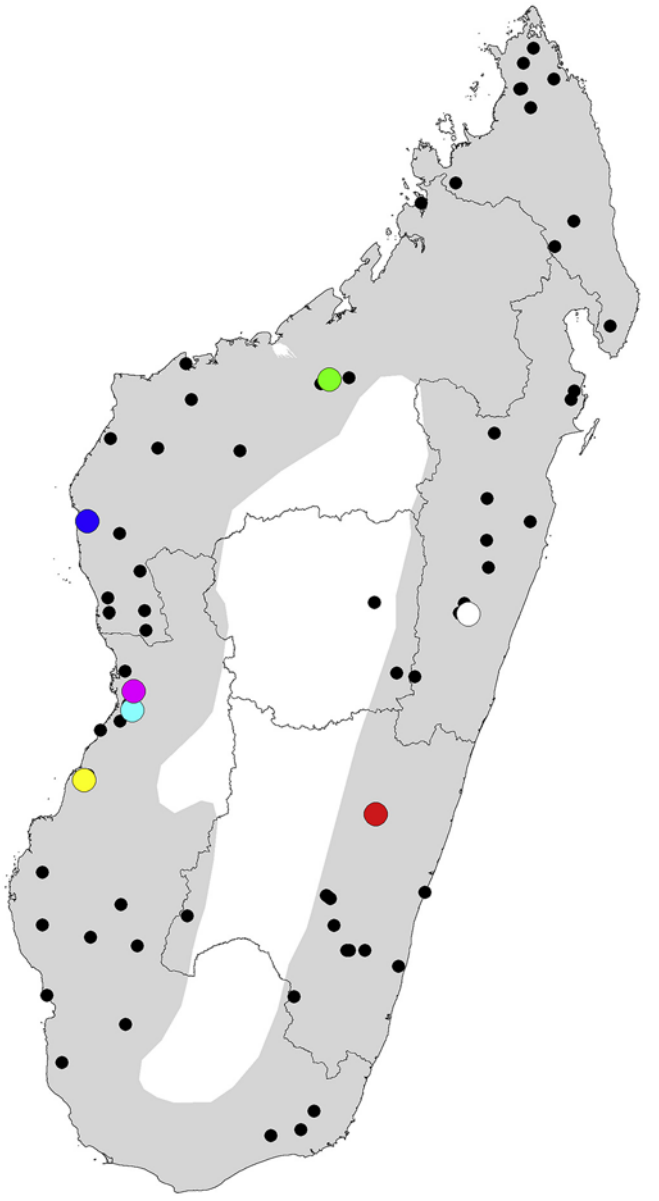

Fig. 1. Distribution of *Cryptoprocta ferox* based on IUCN (2016) in grey, and from Goodman (2013) (dots), and localities of samples (color dots) used in this study. Green: Ankarafantsika; dark blue: Ambinda, Beanka Forest; white: Ambavaniasy; pink: Ambadira; light blue: Kirindy (CNFEREF) Forest; yellow: Kirindy Mitea; red: Ranomafana National Park. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article).

close to the Malagasy mongooses (Galidiinae) or to the other Euplerinae (falanouk and Malagasy civet; see Yoder et al., 2003; Poux et al., 2005). The fosa is the largest living terrestrial predator on the island, sexually dimorphic, with larger males in nature reaching a body mass of over 10 kg. Its diet includes principally mammals, with lemurs often representing the main prey (Rasoloarison et al., 1995; Dollar et al., 2007; Goodman, 2009; Lührs and Dammhahn, 2010; Lührs et al., 2013). Their hunting techniques are linked to particular anatomical features, including large footpads, semi-retractable claws, and flexible ankles (Taylor, 1989; Veron, 1999), allowing them to move and hunt with considerable dexterity both on the ground and in trees.

This species has large home ranges of up to 26 km^2 and daily movements of up to 5–7 km (Dollar, 1999; Hawkins, 2003; Lührs and Kappeler, 2013). Fosas occur at low densities in forested habitats (Hawkins and Racey, 2005; Gerber et al., 2010), to which they are generally restricted, and have the broadest distribution of any Eupleridae (see Fig. 1). The fosa is classified as Vulnerable (IUCN, 2016) because of habitat loss, hunting, other forms of persecution, and the effects of introduced carnivorans (Farris et al., 2015, 2016; IUCN, 2016). Madagascar has indeed undergone a massive reduction of its forest cover over the last decades, and few large blocks remain (Harper et al., 2007; Irwin et al., 2010). Human population growth and socio-economic problems drive reduction and degradation of natural habitat, and wildlife hunting is common (Irwin et al., 2010). Even though some endemic mammal species may have adapted to environmental degradation (e.g., members of the subfamily Tenrecinae), large predators, such as the fosa, are affected by habitat destruction and anthropogenic disturbance.

Molecular studies on the other Malagasy euplerids (Galidiinae species: Bennett et al., 2009; Jansen Van Vuuren et al., 2012; Veron et al., 2017; Euplerinae: *Eupleres goudotii*, Veron and Goodman, 2018) have shown some phylogeographic structure, particularly in *Galidia elegans*. Given that *C. ferox* is a large animal with implicitly higher dispersal capacity, it might be anticipated to exhibit little phylogeographic structure and low genetic polymorphism. However, the genetic structure of this species has not been examined across its range.

Captive breeding programs in zoological parks aim to support the survival of endangered species (Ebenhard, 1995; Gippoliti, 2011), although the ultimate goals and efficiency of such programs have been debated (Snyder et al., 1996; McPhee, 2003; Alroy, 2015). Captive breeding of *Cryptoprocta* started in 1974, and has been notably successful; a total of 316 individuals have been held in captivity and, in 2014, as reported in the studbook, the living population was 136 individuals housed in 57 institutions around the world (Reiter, 2015); more recent unpublished information has the figure at 113 individuals (T. Tetzlaff, pers. comm.). As early as 1954, this species was held in the Smithsonian National Zoological Park (NZP, Washington, D.C., USA), and since 1985, exhibited at the San Diego Zoo (California, USA). In Europe, it has been exhibited in the Basel Zoo (Switzerland) since 1972. The first captive breeding in Europe started in 1974 at the Montpellier Zoo (France) (Albignac, 1975) and in North America in 1989 at San Diego Zoo. To date, only one zoological garden in Asia holds this species (Ueno Zoological Gardens, Tokyo, Japan; Reiter, 2015). The Parc Zoologique et Botanique de Tsimbazaza (PBZT, Antananarivo, Madagascar), has had successful captive breeding since 2011 (Reiter, 2015).

According to the *Cryptoprocta* studbook (Reiter, 2015), wild caught animals have been transferred to zoos around the world, in 1954 and 1966 (NZP), 1967 (San Diego Zoo), 1972 (Basel; Naples, USA), 1973 (Montpellier), 1980 (Johannesburg, South Africa), 1981 (Basel), 1995 (private, then to Duisburg, Germany; Tilburg, Netherlands), 1997 (San Antonio, USA), 1998 (Bester, South Africa), 1999, 2000, and 2009 (PBZT), 2000 (Bester, South Africa; Cedar Creek, USA), 2002 (Omaha, USA), and 2003 (Lubbock, USA). Information on the original geographic origin on Madagascar of these captive animals is unknown, with the exception of those brought to Montpellier Zoo in 1973, which came from the east coast (Albignac, 1975). Exchanges of animals between zoos in Madagascar, Europe, and North America have been conducted to help maintain the genetic diversity of the species (Reiter, 2015). No molecular studies have investigated the genetic diversity of the captive fosa population, specifically the presence of different lineages, which is crucial for the correct management of a captive breeding program.

There are several other reasons for the need of detailed analyses of genetic divergence in populations of *Cryptoprocta*. A larger species, *C. spelea*, is known to have occurred on the island and its presumed extinction is thought to have taken place in the

Table 1
List of the samples included in this study. For each sample, we report: the identification number, the specimen/sample number (AMNH: American Museum of Natural History, New York; FMNH: Field Museum of Natural History, Chicago; ISEM: Institut des Sciences de l'Evolution, Montpellier; MCZ: Harvard Museum of Comparative Zoology, Harvard University, Cambridge; MNHN: Muséum National d'Histoire Naturelle, Paris; NHM: The Natural History Museum, London), the GenBank (Gbk) number, and locality (B: born; Dist.: District; ND: no data; NP: National Park; Prov: Province; Res.: Reserve; SR: Special Reserve; Stb: Studbook number). GenBank numbers in bold represent new sequences produced in this study; others from: Yoder et al. (2003), Flynn et al. (2005), Koepfli et al. (2006), Patou et al. (2009), Hassanin and Veron (2016), and Veron et al. (2017). Identification numbers in bold correspond to samples from dry specimens and tooth.

Species	Identification #	Sample/voucher #	Cytb Gbk #	CR Gbk #	ND2 Gbk #	FGB Gbk #	Locality
Cryptoprocta ferox	CfA110056	AMNH AMCC 110056	KX592625	**MG452211**	**MG452276**	**MG452181**	Ankarafantsika, Ampijoroa, Mahajanga Prov.
Cryptoprocta ferox	CfA110059	AMNH AMCC 110059		**MG452212**			Ankarafantsika, Ampijoroa, Mahajanga Prov.
Cryptoprocta ferox	CfA110062	AMNH AMCC 110062		**MG452213**			Ankarafantsika, Ampijoroa, Mahajanga Prov.
Cryptoprocta ferox	CfA110069	AMNH AMCC 110069		**MG452214**	**MG452277**	**MG452182**	Kirindy Mitea, Ankotrofotsy, Toliara Prov.
Cryptoprocta ferox	CfA110072	AMNH AMCC 110072		**MG452215**	**MG452278**		Kirindy Mitea, Ankotrofotsy, Toliara Prov.
Cryptoprocta ferox	CfA110075	AMNH AMCC 110075		**MG452216**			Kirindy Mitea, Ankotrofotsy, Toliara Prov.
Cryptoprocta ferox	CfA118633	AMNH AMCC 118633		**MG452217**			Kirindy Mitea, Ankotrofotsy, Toliara Prov.
Cryptoprocta ferox	CfA118634	AMNH AMCC 118634		**MG452218**			Kirindy Mitea, Ankotrofotsy, Toliara Prov.
Cryptoprocta ferox	CfA118635	AMNH AMCC 118635	**MG452145**	**MG452219**			Kirindy Mitea, Ankotrofotsy, Toliara Prov.
Cryptoprocta ferox	CfA118636	AMNH AMCC 118636	**MG452146**	**MG452220**	**MG452279**	**MG452183**	Kirindy Mitea, Ankotrofotsy, Toliara Prov.
Cryptoprocta ferox	CfA118637	AMNH AMCC 118637		**MG452221**			Kirindy Mitea, Ankotrofotsy, Toliara Prov.
Cryptoprocta ferox	CfA118638	AMNH AMCC 118638	**MG452147**	**MG452222**			Kirindy Mitea, Ankotrofotsy, Toliara Prov.
Cryptoprocta ferox	CfA118647	AMNH AMCC 118647		**MG452223**	**MG452280**	**MG452184**	Ankarafantsika, Ampijoroa, Mahajanga Prov.
Cryptoprocta ferox	CfA118661	AMNH AMCC 118661		**MG452224**			Ankarafantsika, Ampijoroa, Mahajanga Prov.
Cryptoprocta ferox	CfA118665	AMNH AMCC 118665	**MG452148**	**MG452225**	**MG452281**	**MG452185**	Ankarafantsika, Ampijoroa, Mahajanga Prov.
Cryptoprocta ferox	CfA118666	AMNH AMCC 118666	**MG452149**	**MG452226**	**MG452282**	**MG452186**	Kirindy Mitea, Ankotrofotsy, Toliara Prov.
Cryptoprocta ferox	CfA118668	AMNH AMCC 118668	**MG452150**	**MG452227**		**MG452187**	Kirindy Mitea, Ankotrofotsy, Toliara Prov.
Cryptoprocta ferox	CfA118670	AMNH AMCC 118670	**MG452151**	**MG452228**		**MG452188**	Kirindy Mitea, Ankotrofotsy, Toliara Prov.
Cryptoprocta ferox	CfA118671	AMNH AMCC 118671		**MG452229**			Kirindy Mitea, Ankotrofotsy, Toliara Prov.
Cryptoprocta ferox	CfAY928681	ISIS 027951	AY928681				ND (San Diego Zoo, ISIS 027951, Stb # 0059)
Cryptoprocta ferox	CfC13	MNHN C-13/ISEMT832	**MG452152**	**MG452230**	**MG452283**	FJ391188	ND (Montpellier Zoo, France, captured in East coast, Stb # 0008)
Cryptoprocta ferox	CfCF02	CF02		**MG452231**			Kirindy Forest (CNFEREF), Toliara Prov.
Cryptoprocta ferox	CfCF03	CF03		**MG452232**	**MG452284**	**MG452189**	Kirindy Forest (CNFEREF), Toliara Prov.
Cryptoprocta ferox	CfCF04	CF04		**MG452233**			Kirindy Forest (CNFEREF), Toliara Prov.
Cryptoprocta ferox	CfCF05	CF05		**MG452234**			Kirindy Forest (CNFEREF), Toliara Prov.
Cryptoprocta ferox	CfCF06	CF06		**MG452235**			Kirindy Forest (CNFEREF), Toliara Prov.
Cryptoprocta ferox	CfCF07	CF07		**MG452236**			Kirindy Forest (CNFEREF), Toliara Prov.
Cryptoprocta ferox	CfCF08	CF08		**MG452237**			Kirindy Forest (CNFEREF), Toliara Prov.
Cryptoprocta ferox	CfCF09	CF09		**MG452238**	**MG452285**	**MG452190**	Kirindy Forest (CNFEREF), Toliara Prov.
Cryptoprocta ferox	CfCF10	CF10		**MG452239**			Kirindy Forest (CNFEREF), Toliara Prov.
Cryptoprocta ferox	CfCF11	CF11		**MG452240**	**MG452286**	**MG452191**	Kirindy Forest (CNFEREF), Toliara Prov.
Cryptoprocta ferox	CfCF12	CF12		**MG452241**	**MG452287**	**MG452192**	Kirindy Forest (CNFEREF), Toliara Prov.
Cryptoprocta ferox	CfCF13	CF13		**MG452242**			Kirindy Forest (CNFEREF), Toliara Prov.
Cryptoprocta ferox	CfCF14	CF14	**MG452153**	**MG452243**	**MG452288**	**MG452193**	Kirindy Forest (CNFEREF), Toliara Prov.
Cryptoprocta ferox	CfCF15	CF15	**MG452154**	**MG452244**	**MG452289**		Kirindy Forest (CNFEREF), Toliara Prov.
Cryptoprocta ferox	CfCF16	CF16	**MG452155**	**MG452245**	**MG452290**		Kirindy Forest (CNFEREF), Toliara Prov.
Cryptoprocta ferox	CfCF17	CF17	**MG452156**	**MG452246**	**MG452291**	**MG452194**	Kirindy Forest (CNFEREF), Toliara Prov.
Cryptoprocta ferox	CfCF18	CF18	**MG452157**	**MG452247**	**MG452292**	**MG452195**	Kirindy Forest (CNFEREF), Toliara Prov.
Cryptoprocta ferox	CfCF19	CF19	**MG452158**	**MG452248**			Kirindy Forest (CNFEREF), Toliara Prov.

Cryptoprocta ferox	CfCF20	CF20		**MG452249**			Kirindy Forest (CNFEREF), Toliara Prov.
Cryptoprocta ferox	CfCF22	CF22	**MG452159**	**MG452250**	**MG452293**	**MG452196**	Kirindy Forest (CNFEREF), Toliara Prov.
Cryptoprocta ferox	CfCF23	CF23	**MG452160**				Kirindy Forest (CNFEREF), Toliara Prov.
Cryptoprocta ferox	CfCF24	CF24	**MG452161**	**MG452251**	**MG452294**	**MG452197**	Kirindy Forest (CNFEREF), Toliara Prov.
Cryptoprocta ferox	CfCF25	CF25	**MG452162**	**MG452252**	**MG452295**		Kirindy Forest (CNFEREF), Toliara Prov.
Cryptoprocta ferox	CfCF26	CF26	**MG452163**	**MG452253**			Kirindy Forest (CNFEREF), Toliara Prov.
Cryptoprocta ferox	CfCF27	CF27	**MG452164**	**MG452254**			Kirindy Forest (CNFEREF), Toliara Prov.
Cryptoprocta ferox	CfCF28	CF28		**MG452255**			Kirindy Forest (CNFEREF), Toliara Prov.
Cryptoprocta ferox	CfCF29	CF29	**MG452165**	**MG452256**			Kirindy Forest (CNFEREF), Toliara Prov.
Cryptoprocta ferox	CfCF30	CF30	**MG452166**	**MG452257**			Kirindy Forest (CNFEREF), Toliara Prov.
Cryptoprocta ferox	CfCF31	CF31	**MG452167**	**MG452258**			Kirindy Forest (CNFEREF), Toliara Prov.
Cryptoprocta ferox	CfCF32	CF32	**MG452168**				Kirindy Forest (CNFEREF), Toliara Prov.
Cryptoprocta ferox	CfCF34	CF34	**MG452169**	**MG452259**			Ambadira, Toliara Prov.
Cryptoprocta ferox	CfCH25	CH-25	AY170096		AY170036		ND
Cryptoprocta ferox	**CfM1668**	MNHN 1992-1668	**MG452170**				Ambavaniasy, Toamasina Prov.
Cryptoprocta ferox	CfTC070	MNHN TC070	**MG452171**	**MG452260**	**MG452296**	**MG452198**	ND (Suffolk Wildlife Park, Kessingland, UK, Stb # 0098)
Cryptoprocta ferox	CfTC076	MNHN TC076		**MG452261**			ND (Duisburg Zoo, Germany, parents from Madagascar; Stb # 0046)
Cryptoprocta ferox	CfTC077	MNHN TC077		**MG452262**			ND (Duisburg Zoo, Germany, Stb # 0067)
Cryptoprocta ferox	CfTC078	MNHN TC078		**MG452263**			ND (Duisburg Zoo, Germany, Stb # 0072)
Cryptoprocta ferox	CfTC079	MNHN TC079	**MG452172**	**MG452264**			ND (Duisburg Zoo, Germany, Stb # 0084)
Cryptoprocta ferox	CfTC102	MNHN TC102		**MG452265**			Ranomafana NP, Fianarantsoa Prov.
Cryptoprocta ferox	CfTC103	MNHN TC103		**MG452266**			Ranomafana NP, Fianarantsoa Prov.
Cryptoprocta ferox	**CfTC695**	MNHN TC695	**MG452173**	**MG452267**			Ambinda, Beanka Forest, Mahajanga Prov.
Cryptoprocta ferox	CfTC706	MNHN TC706		**MG452268**		**MG452199**	ND (Duisburg Zoo, Germany, Stb # 0201)
Cryptoprocta ferox	CfTC707	MNHN TC707		**MG452269**		**MG452200**	ND (Duisburg Zoo, Germany, Stb # 0246)
Cryptoprocta ferox	CfTC726	MNHN TC726	**MG452174**	**MG452270**	**MG452297**	**MG452201**	ND (Belfast Zoo, UK, B in Duisburg, Stb # 0086)
Cryptoprocta ferox	CfTC727	MNHN TC727	**MG452175**	**MG452271**	**MG452298**	**MG452202**	ND (Belfast Zoo, UK, B in Kessingland, Stb # 0097)
Cryptoprocta ferox	CfTC728	MNHN TC728	**MG452176**	**MG452272**	**MG452299**	**MG452203**	ND (Artis Zoo, NL, B in Kessingland, Stb # 0095)
Cryptoprocta ferox	CfTC745	MNHN TC745	**MG452177**	**MG452273**		**MG452204**	ND (Parken Zoo, Sweden, B in Duisburg, Stb # 0252)
Cryptoprocta ferox	CfTC746	MNHN TC746	**MG452178**	**MG452274**		**MG452205**	ND (Parken Zoo, Sweden, B in Duisburg, Stb # 0250)
Cryptoprocta ferox	CfTC747	MNHN TC747	**MG452179**			**MG452206**	ND (Parken Zoo, Sweden, B in Duisburg, Stb # 0251)
Cryptoprocta ferox	CfTC760	MNHN TC760	**MG452180**	**MG452275**		**MG452207**	ND (Parc Zoologique de Paris, France, B Duisburg, Stb # 0223)
Eupleres goudotii	EgTC705	MNHN TC705	KX592626		**MG452300**	**MG452208**	
Fossa fossana	FfF156648	FMNH 156648	AY170097		AY170037	**MG452209**	
Galidia elegans	GeF151925	FMNH 151925	KX592627		AY750648		
Galidia elegans	GeF173102	FMNH 173102				KX592659	
Galidictis f. fasciata	GfF156652	FMNH 156652/	AY170100		AY170040	KX592665	
Galidictis f. grandidieri	GgF173158	FMNH 173158	KX592643			KX592671	
Mungotictis decemlineata	MdTC105	MNHN TC105	KX592644			KX592657	
Mungotictis decemlineata	MdTC032	MNHN TC032/C46			KJ419916		
Salanoia concolor	ScTC696	MNHN TC696	KX592653		**MG452301**	**MG452210**	
Urva fusca	Uf	RED2	FJ391215		FJ391231	FJ391191	

past few millennia (Goodman et al., 2004; Crowley, 2010; Meador et al., 2017); however, it is in the realm of possibility that remnant populations are still extant. Further, on the basis of local folk taxonomy, it has been suggested that two forms of *fosa* occur on the island – *fosa mainty* or "black *Cryptoprocta*" and *fosa mena* or "reddish *Cryptoprocta*"; the latter form is said to be smaller than the former (Decary, 1950). Hence, there are aspects that potentially call into question the monotypic taxonomy of living *Cryptoprocta*.

The purpose of this study was, therefore, to 1) evaluate the genetic polymorphism and geographic structure of wild fosa across its natural range, 2) identify potential geographic lineages/conservation units, 3) identify lineages of the captive breeding animals, and 4) evaluate the degree of genetic polymorphism of the captive population. For this purpose, we analyzed genetic diversity of wild and captive individuals of this species using sequences from three mitochondrial and one nuclear markers. These data provide insight into the dispersal capacity of this large predator, or other factors, such as environmental, that may have influenced the geographic structure and patterns of genetic polymorphism in this species. These data may also reveal management units and help to detect the presence of different lineages and genetic polymorphism in captive populations. The results of this study should be useful for the management of *Cryptoprocta* in the wild and captivity.

Material and methods

Sampling, extraction, PCR and sequencing

We analyzed 69 samples (blood, tissues, hair, teeth, and dry tissues from museum specimens) of animals referred to as *Cryptoprocta ferox* (Table 1, Fig. 1). DNA was isolated following a cetyl trimethyl ammonium bromide (CTAB)-based protocol (Winnepenninckx et al., 1993). For museum (dry tissue) and tooth samples, we added dithiothreitol (DTT 1 M, ca 8–15 μL per extract) during tissue lysis to break up disulfide bonds, and we increased the lysis time (up to 72 h).

We sequenced three mitochondrial fragments: Cytochrome b (Cytb), NADH dehydrogenase subunit 2 (ND2), and the hypervariable region 1 of the Control Region (CR), using previously described primers (Cytb: Veron and Heard, 2000; Veron et al., 2004, 2014; ND2: Sorenson et al., 1999; CR: Palomares et al., 2002). To provide an evolutionary assessment independent from mitochondrial markers, we also amplified the nuclear marker Beta-fibrinogen intron 7 (FGB) using primers of Yu and Zhang (2005).

Polymerase chain reactions (PCRs) were performed as in Veron et al. (2014), with annealing temperatures of 50 °C for Cytb and ND2, 61 °C for CR, and 59 °C for FGB. PCR products were sent to Eurofins Genomics (Ebersberg, Germany) for purification and sequencing (on Applied Biosystem® 3730XL DNA Analyzer). Sequences were edited and aligned manually using Bioedit (version 7; Hall, 1999).

Phylogenetic and haplotypic network analyses

Phylogenetic analyses for each marker were performed using Neighbor-Joining (NJ) and Maximum Likelihood (ML), as implemented in MEGA6 (Tamura et al., 2013). We rooted the phylogenetic analyses with representatives of the six other genera of Eupleridae, and one Herpestidae, *Urva fusca*. For ML, the best-fitting model was estimated prior to the analyses using MEGA6, following the Akaike information criterion (AIC). The selected model was implemented in the ML analyses and node robustness was assessed through 1000 bootstrap replicates. Trees were visualized using FigTree 1.4.0 (Rambaut, 2012). We compared resulting topologies and node support; nodes were considered as supported when bootstrap values were ≥70%.

Table 2
Summary of Cytb intraspecific distances within Eupleridae species (this study and Veron et al., 2017).

	Overall mean intrageneric distance	Range of pairwise distances	N
Eupleres	0.4%	0-1.4%	10
Galidia	1.5%	0-3%	12
Galidictis	0.6%	0-1,2%	8
Mungotictis	0.3%	0-2%	56
Salanoia	0.5%	0-1,2%	10
Cryptoprocta	0.6%	0-2%	39

We employed DNAsp 5.10 (Librado and Rosas, 2009) for defining haplotypes. Network (v 4.6, www.fluxus-engineering.com) was used to construct haplotype median-joining networks (Bandelt et al., 1999) for each of the fragments. We computed genetic distances (p-distances within and between groups) and genetic diversity (haplotype and nucleotide diversity) using MEGA6 and DNAsp5.10.

Pedigree of captive breeding populations

We used the information (parents and sex) of the 316 captive *Cryptoprocta ferox* from the 2014 Studbook (Reiter, 2015) to reconstruct the genealogy using Pedigraph 2.4 (Garbe and Da, 2008).

Results

Genetic analyses

A total of 71 individuals (our 69 samples presented herein and two from GenBank, see Table 1) was analyzed for the four fragments. New sequences have been deposited on GenBank (Accession numbers: MG452145 to MG452301). A few samples, particularly teeth and dried tissue from museum specimens, as well as some poorly preserved hair samples, yielded no or partial sequences (see Table 1). Only 17 samples yielded sequences from the three mitochondrial regions, including those from fours zoos and three localities of wild individuals, and only 14 of these yielded sequences for the four fragments. However, the CR fragment was obtained for most samples (65).

The Cytb fragment (length: 1140 bp; number of variable sites: 26; number of parsimony informative sites: 22; n = 39) showed an overall mean distance of 0.6% (see Table 2). The ML tree (model GTR + G + I, Fig. A 1) showed poor resolution and little structure, apart for one clade including captive individuals in different zoos.

The CR fragment (length: 535 bp; number of variable sites: 47; number of parsimony informative sites: 42; n = 65) showed an overall mean distance of 2.1%. The ML tree (model HKY + G; Fig. A 2) was better structured than that derived from Cytb, and composed of two main clades: 1) an animal from Beanka Forest (central west, near Ambinda), one individual from Ranomafana (central east), and some zoo individuals; and 2) all the other wild and zoo individuals.

The ND2 fragment (length: 1044 bp; number of variable sites: 10; number of parsimony informative sites: 9; n = 25) displayed an overall mean distance: 0.2%. The ML tree (model GTR + G + I) showed poor resolution, and only one small group of zoo individuals clustered together.

The FGB fragment (length: 665 bp; number of variable sites: 1; number of parsimony informative sites: 1; n = 28) provided little information, and the only variable site appeared to be heterozygotic.

Table 3
Genetic diversity estimates within Eupleridae species (this study and Veron et al., 2017). N: number of samples; n: number of sites used; h: number of haplotypes; Hd: haplotype diversity, Pi: nucleotide diversity; S: number of polymorphic sites; and k: average number of nucleotide differences. For *Cryptoprocta*, data for Cytb are shown for all individuals, and separately for wild individuals and zoo individuals; and also, the information is provided for a longer fragment (incomplete sequences deleted).

	Cytb							CR							FGB						
	N	n	h	Hd	Pi	S	k	N	n	h	Hd	Pi	S	k	N	n	h	Hd	Pi	S	k
Eupleres	8	321	2	0.2500	0.00078	1	0.250	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Galidia	12	253	5	0.8636	0.01270	7	3.212	13	401	11	0.9744	0.04572	60	18.333	5	338	1	0	0	0	0
Galidictis	8	1118	5	0.7857	0.01035	27	11.571	4	381	2	0.5000	0.00131	1	0.500	8	665	1	0	0	0	0
Mungotictis	56	1125	6	0.5130	0.00124	27	1.394	51	502	19	0.8361	0.01806	39	9.065	46	589	4	0.3903	0.00007	1	0.043
Salanoia	10	248	3	0.7333	0.00565	3	1.400	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Cryptoprocta-all	**39**	**251**	**4**	**0.5857**	**0.00456**	**3**	**1.144**	**65**	**404**	**11**	**0.5890**	**0.01856**	**34**	**7.498**	**28**	**587**	**1**	**0**	**0**	**0**	**0**
Cryptoprocta-wild	**29**	**253**	**4**	**0.3128**	**0.00224**	**4**	**0.567**	**51**	**424**	**7**	**0.3490**	**0.00693**	**24**	**2.444**	**ND**	**ND**	**ND**	**ND**	**ND**	**ND**	**ND**
Cryptoprocta-zoos	**11**	**593**	**3**	**0.6545**	**0.00368**	**5**	**2.182**	**14**	**436**	**5**	**0.7582**	**0.03685**	**34**	**16.066**	**ND**	**ND**	**ND**	**ND**	**ND**	**ND**	**ND**
Cryptoprocta-Cytb-long	36	686	4	0.5317	0.00223	6	1.530														
Cryptoprocta-wild-Cytb-long	27	1129	3	0.2108	0.00159	19	1.801														

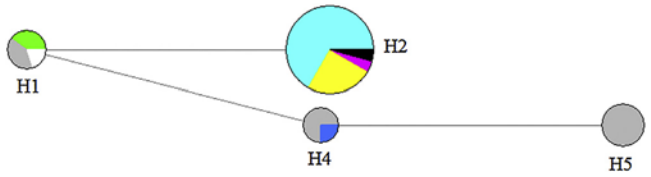

Fig. 2. Median joining network of Cytb haplotypes. The size of each circle is proportional to the haplotype frequency; the shortest link corresponds to one mutation. For color definitions, see Fig. 1.

Cytb haplotype networks were constructed using all sequences, and then only the most complete sequences (longer analyzed fragment). With all sequences (251 bp, n = 39, see Table 3, Fig. 2, Fig. 2), four Cytb haplotypes were obtained, each separated by one to two mutations; H2 grouped wild individuals from several localities in the southwest; H1 included wild individuals from the east and north-west and from zoos in Paris and Montpellier; H4 comprised a wild animal from the central west (Beanka Forest) and zoo individuals, and H3 clustered only captive individuals. When using a longer fragment (686 bp, n = 36), we also obtained four haplotypes, separated by one to three mutations; H2 grouped several localities from the southwest, while other haplotypes each comprised one field locality and zoo samples.

We obtained 11 CR haplotypes (404 bp, n = 65, see Table 3, Fig. 3) separated by one to 32 mutations; they are grouped into several haplogroups. One corresponded to the northwest region (H1, H2, H5, H7), also including samples from zoos in Duisburg, Paris, and Montpellier; one in the southwest region (H3, H4); one in the center-west region (H10), and closely related individuals (H11) from Duisburg zoo (Germany) and Parken zoo (Sweden). The southeast individuals (H8, H9) formed two very distant haplotypes (separated by 19 mutations).

We obtained three ND2 haplotypes (852 bp, n = 25, see Table 3) separated by one to six mutations. The main haplotype, H1, assembled individuals from field localities and one zoo individual, while other haplotypes grouped individuals from zoos.

Measures of polymorphism were calculated for each marker, and separately for wild and zoo individuals (see Table 3). The results showed low haplotype and nucleotide diversity for *C. ferox* (lower than other analyzed euplerids, with the exception of *Mungotictis*), and a comparatively higher genetic diversity in the captive *Cryptoprocta* (based on Cytb, n = 11; and CR, n = 14) as compared to the wild animals (based on Cytb, n = 29; and CR, n = 51).

Within *Cryptoprocta*, the Cytb pairwise distances ranged from 0.0 to 2.0% (Table 2). The Cytb distances between localities ranged from 0.0 to 0.8% (Table A 1). The distances between the four Cytb haplotypes obtained with all sequences (haplotypes based on the 251 bp shared by all sequences) ranged from 0.2 to 0.8% (using the complete Cytb sequences, i.e. 1140 bp). Within *Cryptoprocta*, the CR pairwise distances ranged from 0.0 to 3.0%, and the CR distances between localities ranged from 0.0 to 1.8%.

Genealogy of captive Cryptoprocta

The genealogy included 316 individuals, with information on sex and parentage taken from the studbook (Reiter, 2015). The pedigree obtained was complex, with no isolated lineages. This result is consistent with the numerous exchanges between zoos of breeding *Cryptoprocta* (Fig. A 3; Reiter, 2015).

Discussion

Cryptoprocta ferox is the largest living terrestrial predator on Madagascar, occurring across much of the island (Goodman, 2013), and with considerable dispersal capacity, particularly associated with forest ecosystems (see Lührs, 2012). Hence, as would be anticipated based on these life-history traits and that forest cover was much more extensive on the island until recent historical times, this species, based on samples from across a good portion of its geographic range, including different forest biomes, shows low intraspecific genetic polymorphism (e.g. Cytb: average: 0.6%, ranging from 0.0 to 2.0%, and almost no polymorphism for FGB, which has been shown to vary within carnivoran species, see e.g. Patou et al., 2010; Veron et al., 2015a, 2015b). The measured level of polymorphism was lower than that of another relatively broadly distributed euplerid, *Galidia elegans* (Cytb: average: 1.5%, range: 0–3%, Veron et al., 2017), also sampled in different forest biomes. *Galidia* is distinctly smaller in body size (655–965 g), largely forest-dwelling, and with more limited dispersal capacity than *Cryptoprocta* (Goodman, 2009). The level of genetic polymorphism in *Cryptoprocta* is similar to those measured in the other euplerids (Veron et al., 2017; Veron and Goodman, 2018), but in all cases these taxa are smaller in body size than *Cryptoprocta* and with more restricted geographic ranges.

A comparison of CR results of *Cryptoprocta* and *Mungotictis* samples from the Toliara Province in the central west to southern portions of the island (from this study; Jansen van Vuuren et al., 2012; Veron et al., 2017), illustrates the differences between these two genera. For *Mungotictis*, we found strong polymor-

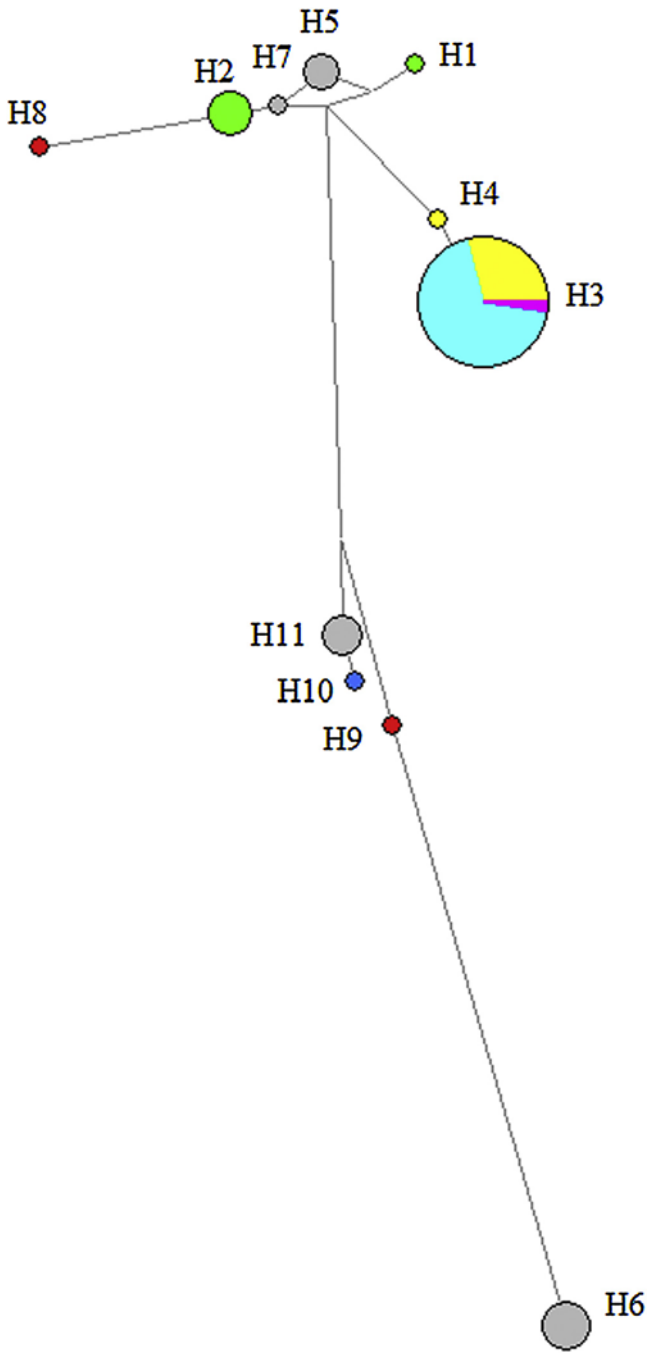

Fig. 3. Median joining network of CR haplotypes. The size of each circle is proportional to the haplotype frequency; the shortest link corresponds to one mutation. For color definitions, see Fig. 1.

phism (19 haplotypes for 51 individuals, and with 36 mutations between the most far apart haplotypes). In contrast, for *Cryptoprocta*, we observed low levels of polymorphism (two haplotypes for 42 individuals, separated only by one mutation; see Table A 2; Fig. A 4). Although this difference could be associated with sampling limitations, it nonetheless indicates important differences in genetic variation patterns between these two monotypic genera.

Mungotictis, which lives in small groups over a vital domain of ca 12–50 ha (see Razafimanantsoa, 2003; Schneider et al., 2016), is strictly forest-dwelling, unknown in open habitats or heavily disturbed forest zones, and has lower dispersal capacity with daily travelled distances of 2200 m (Albignac, 1976; Razafimanantsoa, 2003; Goodman, 2009; Jansen Van Vuuren et al., 2012). In contrast, *Cryptoprocta* is larger, has larger home ranges (up to 26 km^2) and can cross long distances (e.g. 7.3 km in 16 h, Hawkins, 2003), particularly in forest ecosystems, and occasional forays into anthropogenic habitats (Lührs and Kappeler, 2013; Lührs et al., 2013). These aspects might help explain certain inferences associated with dispersal in the fosa and population admixture illustrated by the low mitochondrial DNA polymorphism, and the absence of nuclear DNA polymorphism. The past few decades has seen considerable levels of deforestation in the northern portion of the Toliara Province, resulting in notable fragmentation of the regional forests (Zinner et al., 2014). Given the measured levels of polymorphism in these two genera of euplerids, this would imply that, before habitat fragmentation, *Cryptoprocta* dispersed across the landscape at a distinctly higher rate than *Mungotictis*.

In *Cryptoprocta*, as in smaller euplerids, we obtained some geographic structure, although the genetic distances remained quite low between regions in the fosa. Individuals from the northwest (Ankarafantsika) and east (Ambavaniasy), distinctly different biomes, shared the same Cytb haplotypes. This could be explained by the fact that the elevational range of this genus spans from near sea-level to about 2500 (Goodman, 2013), and, hence, dispersal over the principal north-south aligned mountain range that bisects the island would presumably not pose a barrier, specifically when forest cover was more extensive.

As expected, the sampled populations from the central west, from a limited geographical areas (Ambadira, Kirindy [CNFEREF] Forest, and Kirindy Mitea), were closely related, and shared the same Cytb haplotypes or belong to the same CR haplogroups. Despite the limited geographical distance to the above named populations (from around 220 km to 340 km), the individual from the Beanka Forest, a bit further north, was genetically very distinct (15 mutations between their CR haplotypes; 2 mutations between their Cytb haplotypes). This could be explained by the separation of these areas by the large and meandering Tsiribihina River, which has its headwaters in the eastern portion of the island, and could limit *Cryptoprocta* dispersal between the two regions. Watersheds are known to act as important barriers for other terrestrial vertebrates (see e.g. Goodman and Ganzhorn, 2004; Wilmé et al., 2006). Surprisingly, the two individuals from Ranomafana National Park (center-southeast) were quite distant from each other. These results are difficult to explain, but the genetic data from this site are limited (only CR sequences) due to degraded DNA available from these poorly preserved samples.

One important result of this study is the higher genetic diversity of captive *Cryptoprocta* as compared to wild animals. This can be partially accounted for by certain portions of the island not being represented within the samples used in this study. In turn, the higher genetic diversity of captive *Cryptoprocta* can be explained by the regular sourcing of wild animals into captivity until 2009, based on the studbook (Reiter, 2015). Also, there is good representation of wild lineages found in the current study within zoo populations, as shown by the shared haplotypes between natural populations and animals in zoos. Moreover, we found one zoo lineage that was not represented in the sampled wild populations. Similarly, information from 26 microsatellite loci of 28 fosas from nine European zoos suggested good levels of genetic polymorphism (Vogler et al., 2009).

The genealogy obtained from the studbook provides some background on the parentage of captive born animals, but given

the lack of information on the origin of wild animals brought into captivity, little can be gleaned on the associated phylogeography of the recuperated lineages. The one exception is the individuals sent to the Montpellier zoo in 1973, which were apparently from the northeast coast (Albignac, 1975). These animals were in fact genetically close to wild individuals from the northwest (Ankarafantsika), and not from the northeast as would have been expected based on their origin. However, our results also showed that wild individuals from east and the west (Ankarafantsika and Ambavaniasy) were quite close to each other.

The genetic diversity and presence of different lineages in *Cryptoprocta* held in zoos bodes well for the captive breeding program. Most importantly, data reported herein indicate that further individuals from the wild would not be required for reinforcing or increasing genetic diversity of the zoo populations. As we found no evidence that wild populations contain strongly divergent lineages, we suggest that there is no evidence for and the need to recognize separate conservation units (Manel et al., 2003). As illustrated by our analyses, zoo populations have a strongly mixed pedigree.

Further, based on the absence of strongly divergent lineages, no evidence of extant populations of *C. spelea* was found, nor strong genetic differences that might align with variable morphs or taxa based on folk taxonomy as circumscribed by the *fosa mainty* and *fosa mena* (Decary, 1950).

As the fosa does not commonly occur in human-dominated landscapes (Logan et al., 2015), conservation programs associated with this species should focus on forest habitat protection (see e.g. Kremen et al., 2008) and maintain or reestablish forest corridors linking forest fragments, as proposed for other organisms (see e.g. Schwitzer et al., 2013; Ramiadantsoa et al., 2015). Also, actions should be conducted to reduce human hunting pressure through different types of rural public education programs, and to manage invasive carnivorans that are known to affect the density of native species (Farris et al., 2015, 2016, 2017) and introduce different diseases (Pomerantz et al., 2016; Rasambainarivo and Goodman, in press).

While this current study provides important information on patterns of genetic variability in *Cryptoprocta*, additional genetic studies on wild populations are urgently needed. New samples can be obtained through non-invasive sampling techniques or trapping-releasing wild animals; in the latter case, this needs to be conducted by experienced field staff. In particular, samples are required from zones not covered in the current study. These new analyses should add further insights, specifically to improve geographic coverage, detect other lineages, and conduct finer scale population genetic studies. In particular, it would be crucial to test if habitat fragmentation is affecting the genetic diversity and structure of the species (see e.g. Rivera-Ortiz et al., 2015).

Acknowledgements

We are grateful to the following people for access to specimens and samples, or for their help in the field (see Table 1 for definitions of acronyms): O. Griffiths (Biodiversity Conservation Madagascar), N. Duncan (AMNH, New York, USA), J. Feinstein (AMNH AMCC, New York, USA), J. Chupasko (MCZ, Harvard, USA), A. Kitchener (National Museums Scotland, Edinburgh, UK), S. Langer, J. Reiter and A. Winkler (Zoo Duisburg, Germany), A. Dunham (Rice University), L. Frew (Belfast Zoological Gardens, UK), W. Spencer (Artis Zoo, Amsterdam, NL), A. Akerholm (Parken Zoo, Sweden), A. Lécu (Parc Zoologique de Paris, MNHN, France), F. Catzeflis (ISEM, Université de Montpellier), C. Glaschke (Georg-August-University, Göttingen, Germany), L. Woolaver (Durrell Wildlife Conservation Trust Madagascar), T. Radovimiandrinifarany (Université d'Antananarivo), L.P. Rahajanirina (Friends of Madagascar), B. Andriamihaja (MICET), F.H. Andriamiharisoa, R. Andrianaivoarivelo and J.F. Randrianasolo (FoReT), R. Ampataka, and T. Andrianjanahary.

We warmly thank J. Pfleiderer (Zoo Duisburg) for information on the *Cryptoprocta* studbook, and also L. Lagerström (Parken Zoo, Sweden) and L. Harmon (Naples Zoo, USA) for information on their fosas. We are grateful to A. Hassanin (UMR 7205, MNHN, Paris, France) for his help. Molecular work was undertaken at the 'Service de Systématique Moléculaire' (UMS CNRS 2700, MNHN), and we thank the lab staff for their help, in particular, D. Zuccon and C. Bonillo. GV received financial support from MNHN, CNRS, and from the "Action Transversale du Museum, Biodiversité actuelle et fossile" (MNHN, French Ministry of Research). We are grateful to two anonymous reviewers for their comments on a previous version of this paper.

Appendix A. Supplementary data

Supplementary material related to this article can be found, in the online version, at doi:https://doi.org/10.1016/j.mambio.2018.04.007.

References

Albignac, R., 1975. Breeding the fossa at Montpellier Zoo. Int. Zoo Yearb. 15, 147–150.
Albignac, R., 1976. L'écologie de *Mungotictis decemlineata* dans les forêts décidues de l'ouest de Madagascar. Terre et Vie 30, 347–376.
Alroy, J., 2015. Limits to captive breeding of mammals in zoos. Conserv. Biol 29, 926–931.
Bandelt, H.J., Forster, P., Röhl, A., 1999. Median-joining networks for inferring intraspecific phylogenies. Mol. Biol. Evol. 16, 37–48.
Bennett, C.E., Pastorini, J., Dollar, L., Hahn, W.J., 2009. Phylogeography of the Malagasy ring-tailed mongoose, *Galidia elegans*, from mtDNA sequence analysis. Mitochondrial DNA 20, 7–14.
Crowley, B.E., 2010. A refined chronology of prehistoric Madagascar and the demise of the megafauna. Quat. Sci. Rev. 29, 2591–2603.
Cushman, S.A., Lewis, J., 2010. Movement behavior explains genetic differentiation in American black bears. Landsc. Ecol. 25, 1613–1625.
Decary, R., Payot, Paris 1950. La faune Malgache: son rôle dans les croyances et les usages indigènes.
Dollar, L., 1999. Preliminary report on the status, activity cycle, and ranging of *Cryptoprocta ferox* in the Malagasy rainforest, with implications for conservation. Small Carnivore Conser. 20, 7–10.
Dollar, L., Ganzhorn, J.U., Goodman, S.M., 2007. Primates and other prey in the seasonally variable diet of *Cryptoprocta ferox* in the dry deciduous forest of western Madagascar. In: Gursky, S., Nekaris, K.A. (Eds.), Predation on Primates. Springer, New York, pp. 63–76.
Ebenhard, T., 1995. Conservation breeding as a tool for saving animal species from extinction. Trends Ecol. Evol. 10, 438–443.
Farris, Z.J., Gerber, B.D., Valenta, K., Rafaliarison, R., Razafimahaimodison, J.-C., Larney, E., Rajaonarivelo, T., Randriana, Z., Wright, P.C., Chapman, C.A., 2017. Threats to a rainforest carnivore community: A multi-year assessment of occupancy and co-occurrence in Madagascar. Biol. Conserv. 210, 116–124.
Farris, Z.J., Golden, C.D., Karpanty, S., Murphy, A., Stauffer, D., Ratelolahy, F., Andrianjakarivelo, V., Holmes, C.M., Kelly, M.J., 2015. Hunting, exotic carnivores, and habitat loss: Anthropogenic effects on a native carnivore community, Madagascar. PLoS One 10, http://dx.doi.org/10.1371/journal.pone.0136456.
Farris, Z.J., Kelly, M.J., Karpanty, S., Ratelolahy, F., 2016. Patterns of spatial co-occurrence among native and exotic carnivores in north-eastern Madagascar. Anim. Conserv 19, 189–198.
Flynn, J.J., Finarelli, J.A., Zehr, S., Hsu, J., Nedbal, M.A., 2005. Molecular phylogeny of the Carnivora (Mammalia): Assessing the impact of increased sampling on resolving enigmatic relationships. Syst. Biol 54, 317–337.
Garbe, J.R., Da, Y., 2008. Pedigraph: a software tool for the graphing and analysis of large complex pedigree. In: User manual Version 2.4. University of Minnesota: Department of Animal Science.
Gerber, B., Karpanty, S.M., Crawford, C., Kotschwar, M., Randianantenaina, J., 2010. An assessment of carnivore relative abundance and density in the eastern rainforests of Madagascar using remotely-triggered camera traps. Oryx 44, 219–222.
Gippoliti, S., 2011. Zoos and conservation in the XXI Century: overlooked meeting points between ecology and social sciences? Museologia Scientifica 5, 168–176.

Goodman, S.M., 2009. Family Eupleridae. Madagascar carnivores. In: Wilson, D., Mittermeier, R.A. (Eds.), Handbook of the Mammals of the World, vol. 1. Lynx edicions, Barcelona, pp. 330–351, Carnivores.
Goodman, S.M., 2013. Carnivorans or the order Carnivora. In: Goodman, S.M., Raherilalao, M.J. (Eds.), Atlas of Selected Land Vertebrates of Madagascar. Association Vahatra, Antananarivo, pp. 271–285.
Goodman, S.M., Ganzhorn, J.U., 2004. Biogeography of lemurs in the humid forests of Madagascar: the role of elevational distribution and rivers. J. Biogeogr. 1, 47–55.
Goodman, S.M., Rasoloarison, R.M., Ganzhorn, J.U., 2004. On the specific identification of subfossil *Cryptoprocta* (Mammalia, Carnivora) from Madagascar. Zoosystema 26, 129–143.
Hall, T.E., 1999. BioEdit: a user-friendly biological sequence alignment editor and analysis program for Windows 95/98/NT. Nucl. Acids S 41, 95–98.
Harper, G.J., Steininger, M.K., Tucker, C.J., Juhn, D., Hawkins, F., 2007. Fifty years of deforestation and forest fragmentation in Madagascar. Environ. Conserv 34, 325–333.
Hassanin, A., Veron, G., 2016. The complete mitochondrial genome of the boky-boky, *Mungotictis decemlineata*, the first representative of the Malagasy carnivores (Mammalia, Carnivora, Eupleridae). Mitochondrial DNA 27, 908–909.
Hawkins, C.E., 2003. *Cryptoprocta ferox*, Fossa, Fosa. In: Goodman, S.M., Benstead, J.P. (Eds.), The Natural History of Madagascar. The University of Chicago Press, Chicago, pp. 1360–1363.
Hawkins, C.E., Racey, P.A., 2005. Low population density of a tropical forest carnivore, *Cryptoprocta ferox*: Implications for protected area management. Oryx 39, 35–43.
IUCN, e.T5760A97189 2016. The IUCN Red List of Threatened Species 2016., http://dx.doi.org/10.2305/IUCN.UK.2016-1.RLTS.T5760A97189.en (accessed 3 May 2017).
Irwin, M.T., Wright, P.C., Birkinshaw, C., Fisher, B.L., Gardner, C.J., Glos, J., Goodman, S.M., Loiselle, P., Robeson, P., Raharison, J.L., Raherilalao, M.J., Rakotondravony, D., Raselimanana, A., Ratsimbazafy, J., Sparks, J.S., Wilmé, L., Ganzhorn, J.U., 2010. Patterns of species change in anthropogenically disturbed forests of Madagascar. Biol. Conserv. 143, 2351–2362.
Jansen Van Vuuren, B., Woolaver, L., Goodman, S.M., 2012. Genetic population structure in the boky-boky (Carnivora: Eupleridae), a conservation flagship species in the dry deciduous forests of central western Madagascar. Anim. Conserv. 15, 164–173.
Koepfli, K.P., Jenks, S.M., Eizirik, E., Zahirpour, T., Van Valkenburgh, B., Wayne, R.K., 2006. Molecular systematics of the Hyaenidae: relationships of a relictual lineage resolved by a molecular supermatrix. Mol. Phylogenet. Evol. 38, 603–620.
Kremen, C., Cameron, A., Moilanen, A., Phillips, S.J., Thomas, C.D., Beentje, H., Dransfield, J., Fisher, B.L., Glaw, F., Good, T.C., Harper, G.J., Hijmans, R.J., Lees, D.C., Louis Jr., E., Nussbaum, R.A., Raxworthy, C.J., Razafimpahanana, A., Schatz, G.E., Vences, M., Vieites, D.R., Wright, P.C., Zjhra, M.L., 2008. Aligning conservation priorities across taxa in Madagascar with high-resolution planning tools. Science 320 (5873), 222–226, http://dx.doi.org/10.1126/science.1155193.
Librado, P., Rosas, J., 2009. DnaSP v5: A software for comprehensive analysis of DNA polymorphism data. Bioinformatics 25, 1451–1452.
Logan, M.K., Gerber, B.D., Karpanty, S.M., Justin, S., Rabenahy, F.N., 2015. Assessing carnivore distribution from local knowledge across a human-dominated landscape in central-southeastern Madagascar. Anim. Conserv. 18, 82–91.
Lührs, M.L., 2012. Social organisation and mating system of the fosa (*Cryptoprocta ferox*). PhD Thesis. Georg-August-Universität Göttingen.
Lührs, M.L., Dammhahn, M., 2010. An unusual case of cooperative hunting in a solitary carnivore. J. Ethol. 28, 379–383.
Lührs, M.L., Kappeler, P.M., 2013. Simultaneous GPS tracking reveals male associations in a solitary carnivore. Behav. Ecol. Sociobiol. 67, 1731–1743.
Lührs, M.L., Dammhahn, M., Kappeler, P.M., 2013. Strength in numbers: Males in a carnivore grow bigger when they associate and hunt cooperatively. Behav. Ecol. 24, 21–28.
Manel, S., Schwartz, M.K., Luikart, G., Taberlet, P., 2003. Landscape genetics: combining landscape ecology and population genetics. Trends Ecol. Evol. 18, 189–197.
McPhee, M.E., 2003. Generations in captivity increases behavioral variance: considerations for captive breeding and reintroduction programs. Biol. Conserv 115, 71–77.
Meador, L.R., Godfrey, L.R., Rakotondramavo, J.C., Ranivoharimanana, L., Zamora, A., Sutherland, M.R., Irwin, M.T., 2017. *Cryptoprocta spelea* (Carnivora: Eupleridae): What did it eat and how do we know? J. Mamm. Evol., http://dx.doi.org/10.1007/s10914-017-9391-z.
Palomares, F., Godoy, J., Piriz, A., O'Brien, S., Johnson, W., 2002. Faecal genetic analysis to determine the presence and distribution of elusive carnivores: design and feasibility for the Iberian lynx. Mol. Ecol. 11, 2171–2182.
Patou, M.L., McLenachan, P.A., Morley, C.G., Couloux, A., Jennings, A.P., Veron, G., 2009. Molecular phylogeny of the Herpestidae (Mammalia, Carnivora) with a special emphasis on the Asian Herpestes. Mol. Phylogenet. Evol. 53, 69–80.
Patou, M.L., Wilting, A., Gaubert, P., Esselstyn, J.A., Cruaud, C., Jennings, A.P., Fickel, J., Veron, G., 2010. Evolutionary history of the *Paradoxurus* palm civets – a new model for Asian biogeography. J. Biogeogr. 37, 2077–2097.
Pomerantz, J., Rasambainarivo, F.T., Dollar, L., Rahajanirina, L.P., Andrianaivoarivelo, R., Parker, P., Dubovi, E., 2016. Prevalence of antibodies to selected viruses and parasites in introduced and endemic carnivores in western Madagascar. J. Wildl. Dis. 52, 544–552.
Poux, C., Madsen, O., Marquard, E., Vieites, D.R., de Jong, W.W., Vences, M., 2005. Asynchronous colonization of Madagascar by the four endemic clades of primates, tenrecs, carnivores, and rodents as inferred from nuclear genes. Syst. Biol. 54, 719–730.
Rambaut, A., 2012. Figtree Version 1.4.0 (accessed 30 September 2013) http://tree.bio.ed.ac.uk/software/figtree/.
Ramiadantsoa, T., Ovaskainen, O., Rybicki, J., Hanski, I., 2015. Large-scale habitat corridors for biodiversity conservation: a forest corridor in Madagascar. PLoS One 10 (7), e0132126, http://dx.doi.org/10.1371/journal.pone.0132126.
Rasambainarivo, F.T., Goodman, S.M., 2018. Diseases risks from introduced species to malagasy wildlife. In: Miller, R.E., Calle, P., Lamberski, N. (Eds.), Fowler'S Zoo and Wildlife Medicine. , 9th edition. W. B. Saunders, St. Louis.
Rasoloarison, R.M., Rasolonandrasana, B.P.N., Ganzhorn, J.U., Goodman, S.M., 1995. Predation on vertebrates in the Kirindy Forest, western Madagascar. Ecotropica 1, 59–65.
Razafimanantsoa, L., 2003. *Mungotictis decemlineata*, narrow-striped mongoose, *boky-boky*. In: Goodman, S.M., Benstead, J.P. (Eds.), The Natural History of Madagascar. The University of Chicago Press, Chicago, pp. 1357–1360.
Reiter, J., 2015. Fossa *Cryptoprocta ferox* International Studbook 2014. EAZA, Zoo Duisburg, Duisburg.
Rivera-Ortiz, F.A., Aguilar, R., Arizmendi, M.D.C., Quesada, M., Oyama, K., 2015. Habitat fragmentation and genetic variability of tetrapod populations. Anim. Conserv. 18, 249–258.
Schneider, T.C., Kappeler, P.M., Pozzi, L., 2016. Genetic population structure and relatedness in the narrow-striped mongoose (*Mungotictis decemlineata*), a social Malagasy carnivore with sexual segregation. Ecol. Evol. 11, 3734–3749.
Schwitzer, C., Mittermeier, R., Davies, N., Johnson, S., Ratsimbazafy, J., Razafindramanana, J., Louis Jr., E.E., Rajaobelina, S., 2013. Lemurs of Madagascar: A Strategy for Their Conservation 2013–2016. IUCN SSC Primate Specialist Group. Bristol Conservation and Science Foundation, and Conservation International, Bristol.
Snyder, N.F.R., Derrickson, S.R., Beissinger, S.R., Wiley, J.W., Smith, T.B., Toone, W.D., Miller, B., 1996. Limitations of captive breeding in endangered species recovery. Conserv. Biol. 10, 338–348.
Sorenson, M.D., Ast, J.C., Dimcheff, D.E., Yuri, T., Mindell, D.P., 1999. Primers for PCR-based approach to mitochondrial genome sequencing in birds and other vertebrates. Mol. Phylogenet. Evol. 12, 105–114.
Tamura, K., Stecher, G., Peterson, D., Filipski, A., Kumar, S., 2013. MEGA6: Molecular Evolutionary Genetics Analysis version 6.0. Mol. Biol. Evol. 30, 2725–2729.
Taylor, M.E., 1989. Locomotor adaptations by carnivores. In: Gittleman, J.L. (Ed.), Carnivore Behaviour, Ecology and Evolution. Chapman and Hall, London, pp. 382–409.
Veron, G., 1999. Pads morphology in the Viverridae (Carnivora). Acta Theriol 44, 363–376.
Veron, G., Dupré, D., Jennings, A.P., Gardner, L.C.J., Hassanin, A., Goodman, S.M., 2017. New insights into the systematics of Malagasy mongoose-like carnivorans (Carnivora, Eupleridae, Galidiinae) based on mitochondrial and nuclear DNA sequences. J. Zool. Syst. Evol. Res. 55, 250–264.
Veron, G., Goodman, S.M., 2018. One or two species of the rare Malagasy carnivoran *Eupleres* (Eupleridae)? New insights from molecular data. Mammalia 2, 107–112, http://dx.doi.org/10.1515/mammalia-2016-0182.
Veron, G., Heard, S., 2000. Molecular systematics of the Asiatic Viverridae (Carnivora) inferred from mitochondrial cytochrome b sequence analysis. J. Zool. Syst. Evol. Res. 38, 209–217.
Veron, G., Heard Rosenthal, S., Long, B., Roberton, S., 2004. The molecular systematics and conservation of an endangered carnivore, the Owston's palm civet *Chrotogale owstoni* (Thomas, 1912) (Carnivora, Viverridae, Hemigalinae). Anim. Conserv 7, 107–112.
Veron, G., Patou, M.L., Debruyne, R., Couloux, A., Fernandez, D.A.P., Wong, S.T., Fuchs, J., Jennings, A.P., 2015a. Systematics of the Southeast Asian mongooses (Herpestidae, Carnivora) – Solving the mystery of the elusive collared mongoose and Palawan mongoose. Zool. J. Linn. Soc. 173, 236–248.
Veron, G., Patou, M.L., Jennings, A.P., 2015b. Molecular systematics of the small-toothed palm civet (*Arctogalidia trivirgata*) reveals a strong divergence of Bornean populations. Mamm. Biol 80, 347–354.
Veron, G., Willsch, M., Dacosta, V., Patou, M.L., Seymour, A., Bonillo, C., Couloux, A., Wong, S.T., Jennings, A.P., Fickel, J., Wilting, A., 2014. The distribution of the Malay civet *Viverra tangalunga* (Carnivora: Viverridae) across Southeast Asia: natural or human-mediated dispersal? Zool. J. Linn. Soc. 170, 917–932.
Vogler, B.R., Bailey, C.A., Shore, G.D., McGuire, S.M., Engberg, S.E., Fickel, J., Louis Jr., E.E., Brenneman, R.A., 2009. Characterization of 26 microsatellite marker loci in the fosa (*Cryptoprocta ferox*). Conserv. Genet. 10, 1449–1453.
Whitmee, S., Orme, C.D.L., 2013. Predicting dispersal distance in mammals: a trait-based approach. J. Anim. Ecol. 82, 211–221.
Wilmé, L., Goodman, S.M., Ganzhorn, J.U., 2006. Biogeographic evolution of Madagascar's microendemic biota. Science 312, 1063–1065.
Winnepenninckx, B., Backeljau, T., De Wachter, R., 1993. Extraction of high molecular weight DNA from molluscs. Trends Genet 9, 407.
Yoder, A.D., Burns, M.M., Zehr, S., Delefosse, T., Veron, G., Goodman, S.M., Flynn, J.J., 2003. Single origin of Malagasy Carnivora from an African ancestor. Nature 421, 734–737.

Yu, L., Zhang, Y.P., 2005. Phylogenetic studies of pantherine cats (Felidae) based on multiple genes, with novel application of nuclear beta-fibrinogen intron 7 to carnivores. Mol. Phylogenet. Evol. 35, 483–495.

Zinner, D., Wygoda, C., Razafimanantsoa, L., Rasoloarison, R., Andrianandrasana, H.T., Ganzhorn, J.U., Torkler, F., 2014. Analysis of deforestation patterns in the central Menabe, Madagascar, between 1973 and 2010. Reg. Environ. Change 14, 157–166.